EPIDERMIS
CORTEX
ENDODERMIS
PERICYCLE
LATERAL ROOT CAP
COLUMELLA ROOT CAP
VASCULAR TISSUE
CORTEX/ENDODERMAL INITIAL
EPIDERMAL/ROOT CAP INITIAL
QUIESCENT CENTER
COLUMELLA ROOT CAP INITIAL

PERICLINAL DIVISION

ANTICLINAL DIVISION

C E
C E In
In

Da
In In

FIG. 1A

FIG. 1B

FIG. 1C

FIG. 1D

FIG. 1E

FIG. 1F

FIG. 1G

FIG. 1H

FIG. 2A

```
LS   144   PFIRFTQLTANQAILEAINGNHQAIHIVDFDINHGVQWPPLMQALADRYPA-PTLRITG
GAI  248   PYLKFAHFTANQAILEAFQGK-KRVHVIDFSMSQGLQWPALMQALALRPGGPPVFRLTG
RGA  301   PYLKFAHFTANQAILEAFEGK-KRVHVIDFSMNQGLQWPALMQALALREGGPPTFRLTG
SCR  379   PLVKFSHFTANQAIQEAFEKE-DSVHIIDLDIMQGLQWPGLFHILASRPGGPPHVRLTG
SHR  233   PWATFGHVAANGAILEAVDGE-AKIHIVDISSTFCTQWPTLLEALATRSDDTPHLRLTT
```

FIG. 2B

FIG. 2C

WT   *shr-1*   *shr-2*   *shr-3*   *scr-1*

SHR
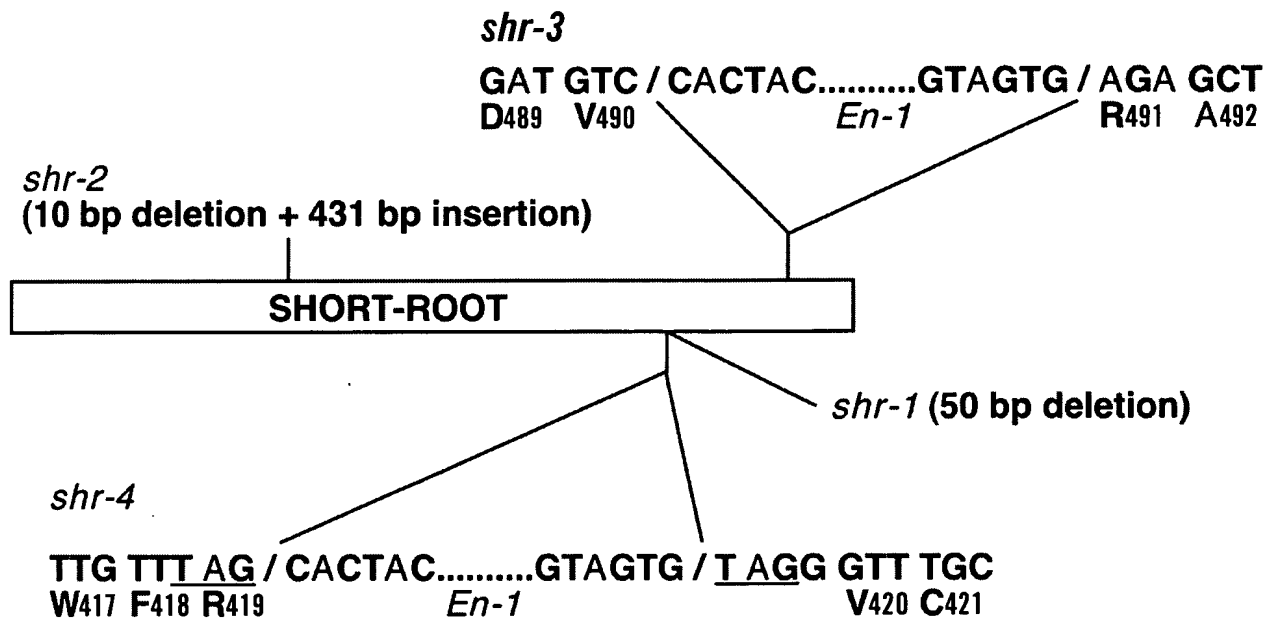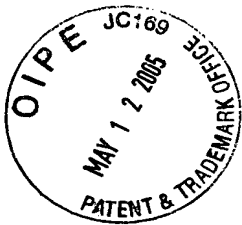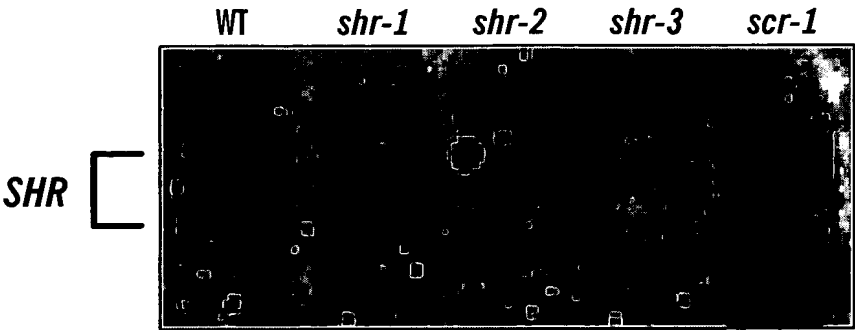
FIG. 3A
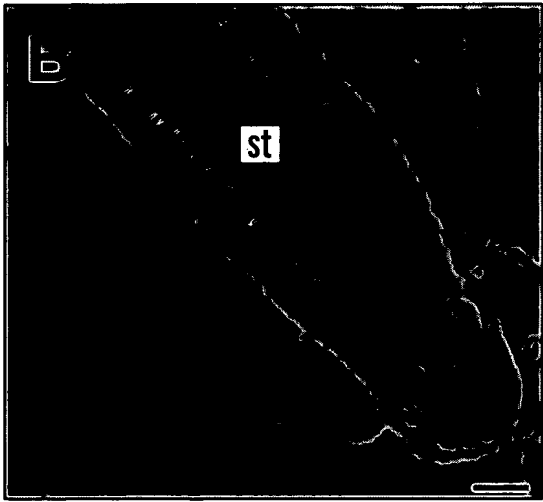
FIG. 3B   *FIG. 3C*
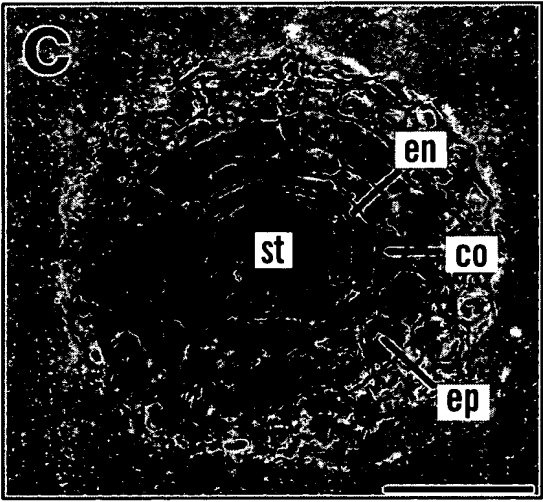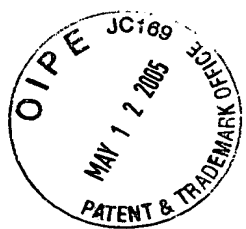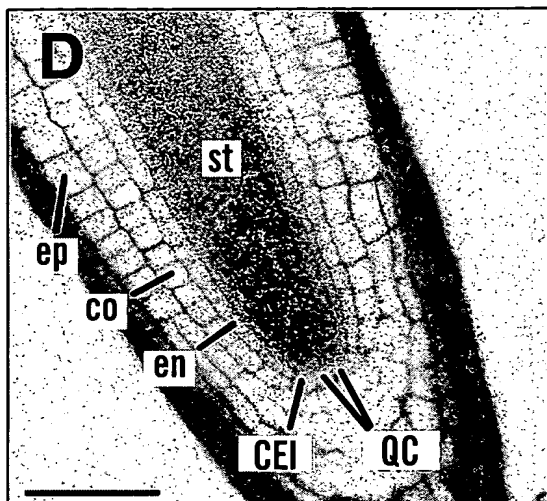
FIG. 3D
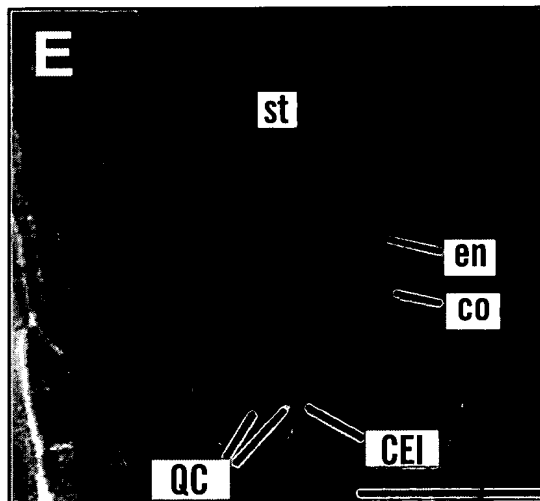
FIG. 3E
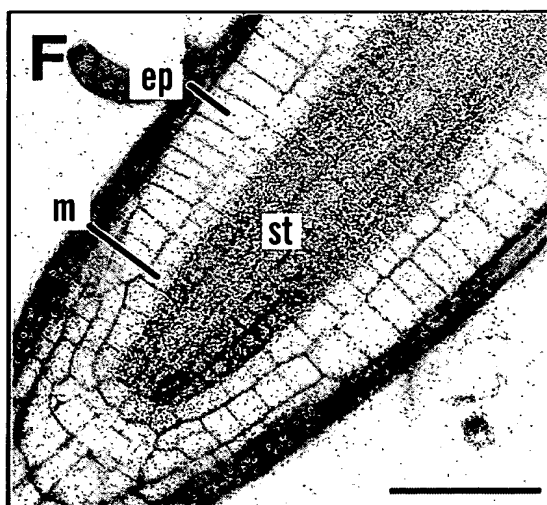
FIG. 3F
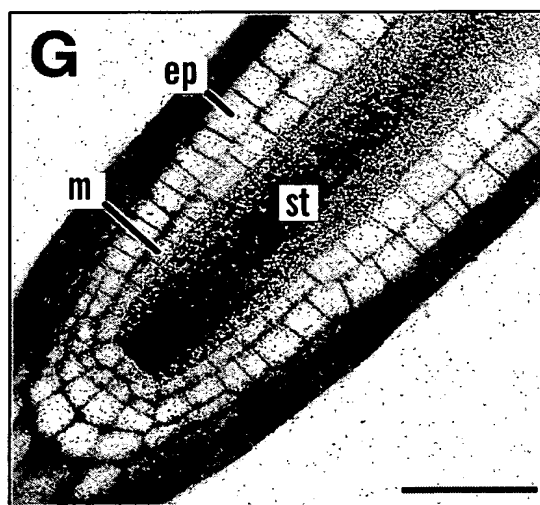
FIG. 3G
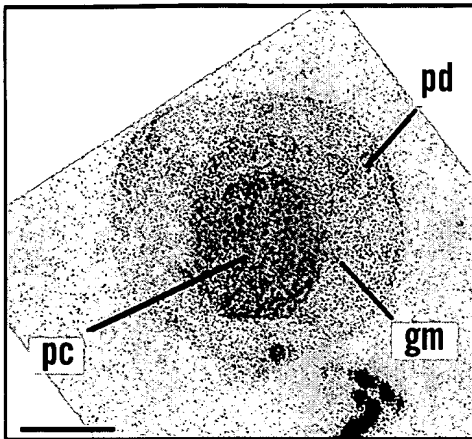
FIG. 4A

FIG. 4B
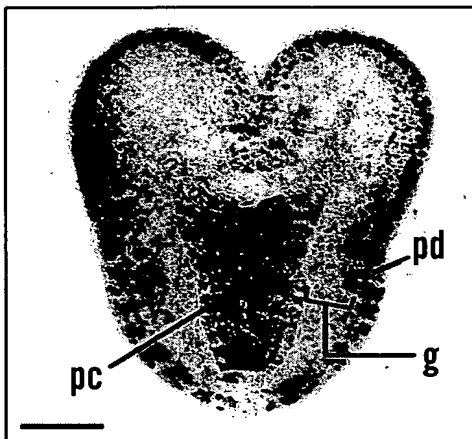
FIG. 4C

FIG. 4D

FIG. 4E

FIG. 4F

WT   shr-1   shr-2

SCR

GDH1

FIG. 5A

st
ep
co
en
d
QC   CEI
WT

FIG. 5B

st
ep
m
shr-2
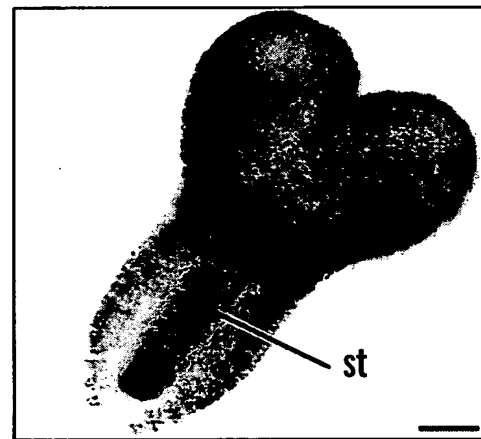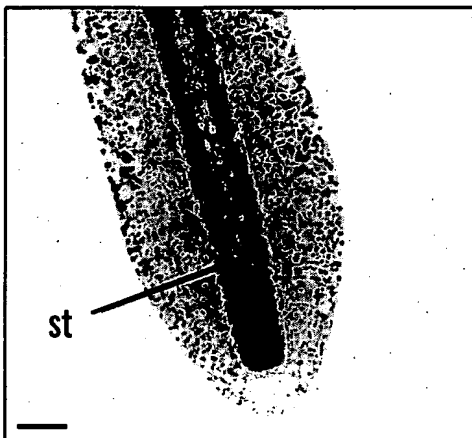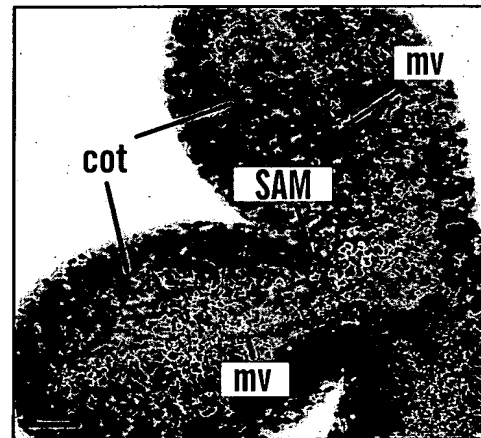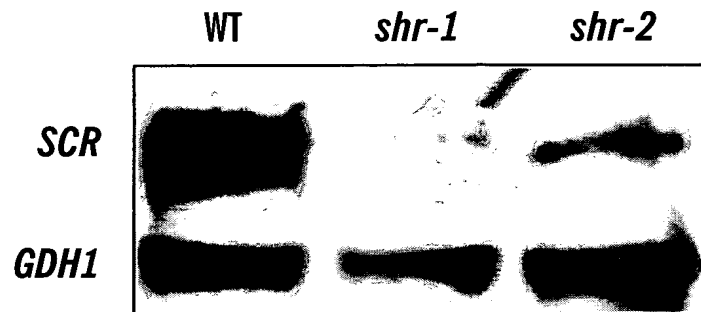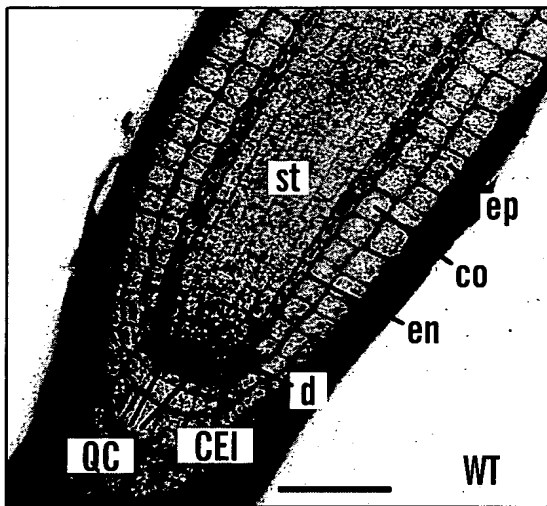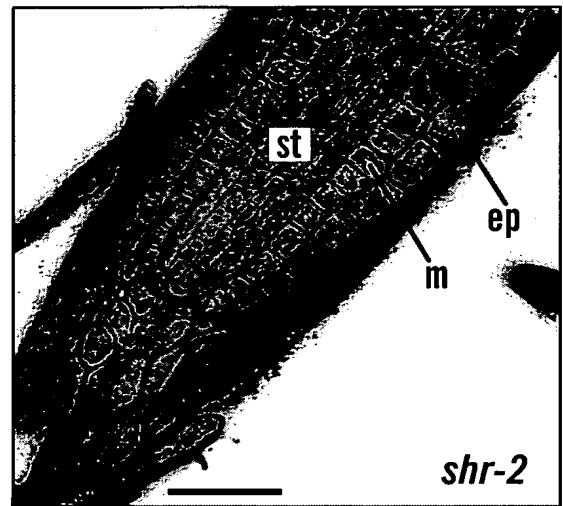
FIG. 5C
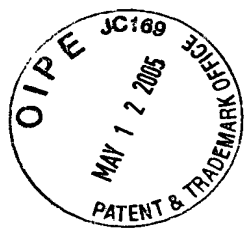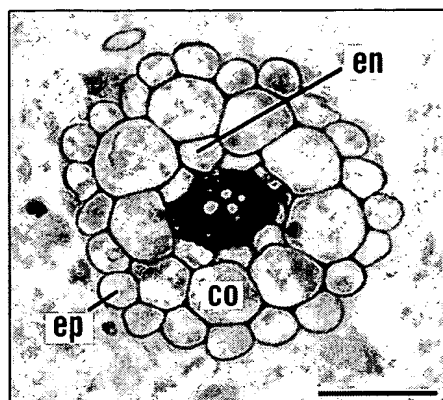
FIG. 6A
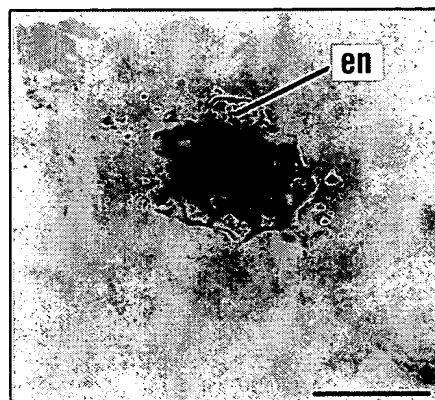
FIG. 6B
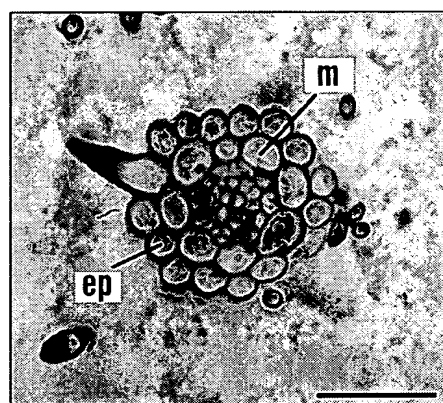
FIG. 6C
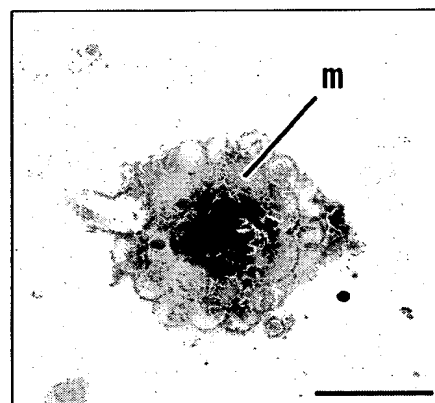
FIG. 6D
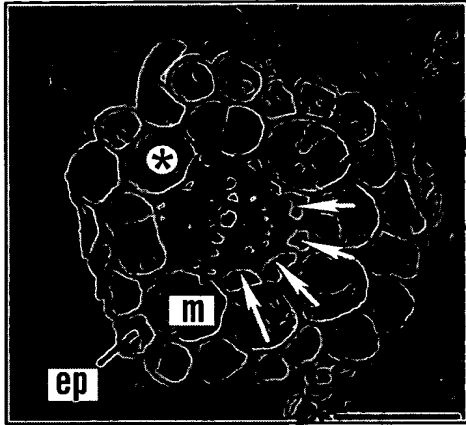
FIG. 6E
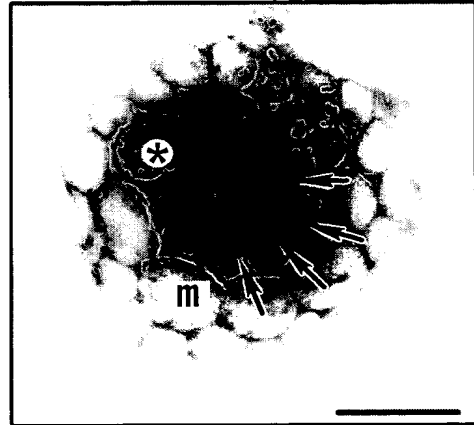
FIG. 6F

FIG. 6G

FIG. 6H

FIG. 7A
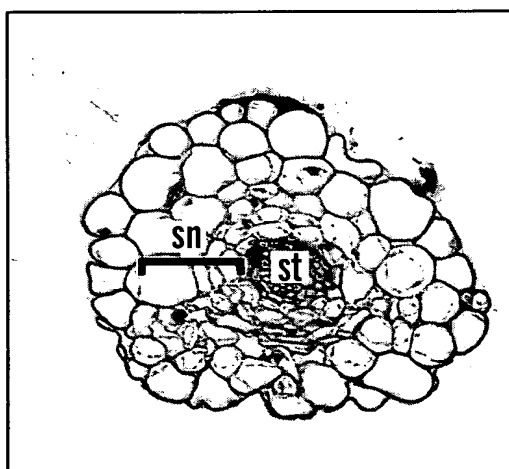
FIG. 7B
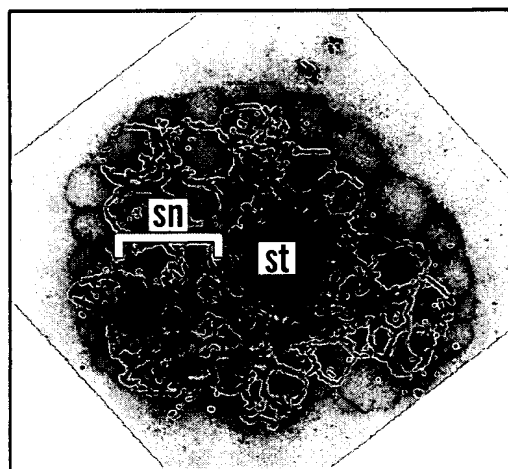
FIG. 7C
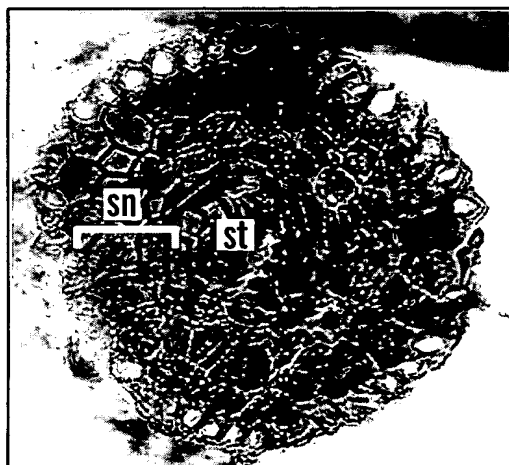
FIG. 7D
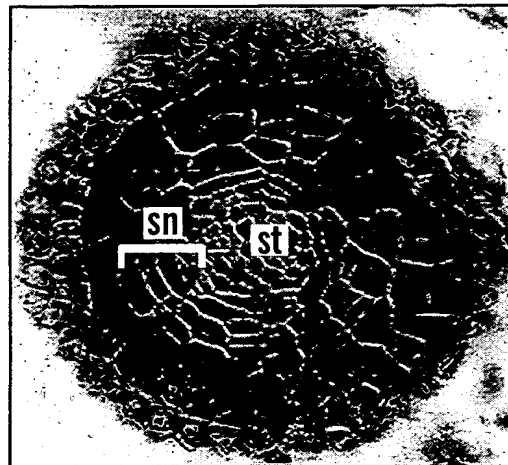
FIG. 7E
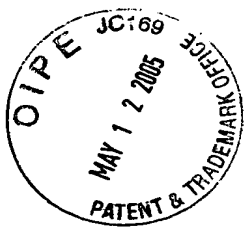
```
   1 atcgattaag agaaaataga gttttcatgc accagtgttg atagtaacgt agtcgcggaa
  61 tgtctaaaac gattatgagt ttggtgtttt gattggttag aattggtatt agtaggacat
 121 tctaactttt ttgttagtct gttgatttag gatgcgtaaa gagtcttttt attttacacc
 181 agttgagact tgggatcgat agtacttgaa acacttggtt ggtttcatgt atttggccta
 241 tatataaaca aacatcgtaa ttatatacgg attttttcg gaattttacg ccatatctgt
 301 aagtatatat aacatgcatg tcgttttcaa attcatatga tgaacgatcc acgtaagtgc
 361 tactactcct acaatattgc atgagagaga tatgtattta taaattttat tttgaagaag
 421 aaataagagg gaaggttact tgggtggatc gatgtgaaaa caaaagaaga aaaagcgaaa
 481 cccactaagc cattacatga tatcgacctt cttatctttt tcctctttat tttatttttc
 541 tcaggacttt tttctactta atgaaacctc caaactatct aactaataca ctcccatgta
 601 gaataaagaa aattatataa gatattgttg atatttgta actagaaaat atatttgctc
 661 tgtaattttt cgtaagttaa atcaacattt ttcagtagaa acaaatatta ctgcaaaaag
 721 taggatcatt attttgtcc aaaatctcag ttagctatag ggttgtagta aaaacaaaac
 781 acattcttga tttgccccaa aaaataaaga gagagaagaa tattgttcaa aagtggtctc
 841 ttctctctct aattatgttt tcactaaacc caattagatt caaacagtct acaaagtcca
 901 aaagataaac atgggacaac aattcgatgc aaaaaatcct cttttcatgc tcttttttta
 961 ttctctagtc ttttaaatta ctaataaaaa ctcacaaatc caccaaaccc attctctaca
1021 actcaccttc atctagattt acccactccc accgagaaac acaagaaaaa aaatatacat
1081 atataaatat acaagacaac acatgatgct gatgcaatat acacaacaaa gtattaaatc
1141 ttagatattg tgggtctccc tttcttctat tcattttctt attcattaaa aaaaaaaaat
1201 ggatactctc tttagactag tcagtctcca acaacaacaa caatccgata gtatcattac
1261 aaatcaatct tcgttaagca gaacttccac caccactact ggctctccac aaactgctta
1321 tcactacaac tttccacaaa acgacgtcgt cgaagaatgc ttcaacttttt tcatggatga
1381 agaagacctt tcctcttctt cttctcacca caaccatcac aaccacaaca atcctaatac
1441 ttactactct cctttcacta ctcccaccca ataccatccc gccacatcat caaccccttc
1501 ctccaccgcc gcagccgcag ctttagcctc gccttactcc tcctccggcc accataatga
1561 cccttccgcg ttctccatac ctcaaactcc tccgtccttc gacttctcag ccaatgccaa
1621 gtgggcagac tcggtccttc ttgaagcggc acgtgccttc tccgacaaag acactgcacg
1681 tgcgcaacaa atcctatgga cgctcaacga gctctcttct ccgtacggag acaccgagca
1741 aaaactggct tcttacttcc tccaagctct cttcaaccgc atgaccggtt caggcgaacg
1801 atgctaccga accatggtaa cagctgcagc cacagagaag acttgctcct tcgagtcaac
1861 gcgaaaaact gtactaaagt tccaagaagt tagcccctgg gccacgtttg gacacgtggc
1921 ggcaaacgga gcaatcttgg aagcagtaga cggagaggca aagatccaca tcgttgacat
1981 aagctccacg ttttgcactc aatggccgac tcttctagaa gctttagcca caagatcaga
2041 cgacacgcct cacctaaggc taaccacagt tgtcgtggcc aacaagtttg tcaacgatca
2101 aacggcgtcg catcggatga tgaaagagat cggaaaccga atggagaaat tcgctaggct
2161 tatgggagtt cctttcaaat ttaacattat tcatcacgtt ggagatttat ctgagtttga
2221 tctcaacgaa ctcgacgtta aaccagacga agtcttggcc attaactgcg taggcgcgat
2281 gcatgggatc gcttcacgtg gaagccctag agacgctgtg atatcgagtt ccgacggtt
2341 aagaccgagg attgtgacgg tcgtagaaga agaagctgat cttgtcggag aagaagaagg
2401 tggctttgat gatgagttct tgagagggtt tggagaatgt ttacgatggt ttagggtttg
2461 cttcgagtca tgggaagaga gttttccaag gacgagcaac gagaggttga tgctagagcg
2521 tgcagcggga cgtgcgatcg ttgatcttgt ggcttgtgag ccgtcggatt ccacggagag
2581 gcgagagaca gcgaggaagt ggtcgaggag gatgaggaat agtgggtttg gagcggtggg
2641 gtatagtgat gaggtggcgg atgatgtcag agctttgttg aggagatata aagaaggtgt
2701 ttggtcgatg gtacagtgtc ctgatgccgc cggaatattc ctttgttgga gagatcagcc
2761 ggtggtttgg gctagtgcgt ggcggccaac gtaaagggtt gtttttattt tttcataagg
2821 aattc
```
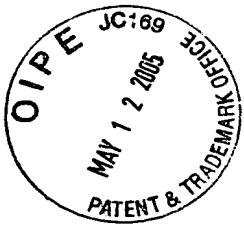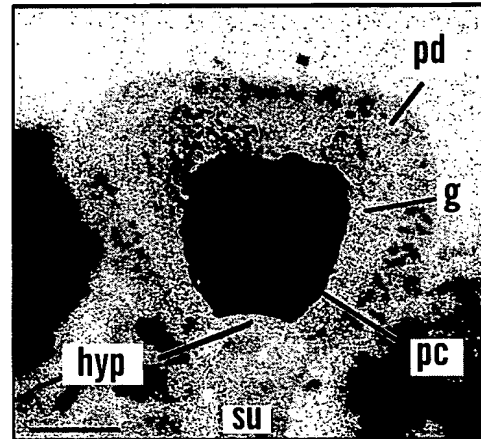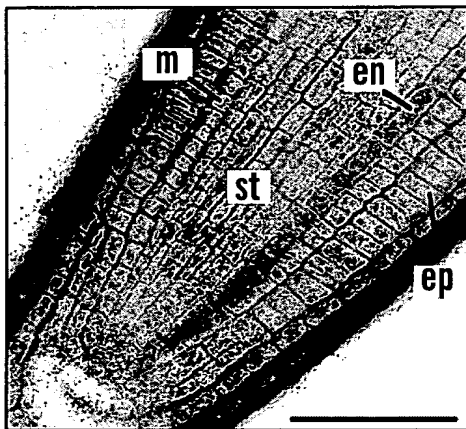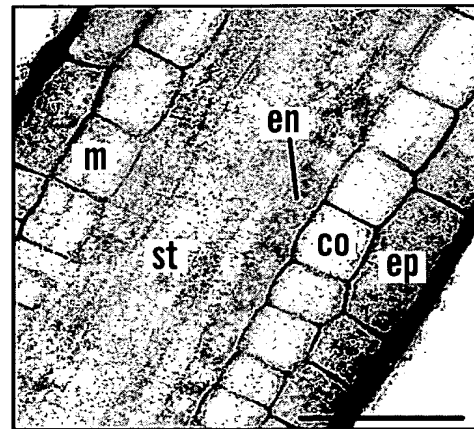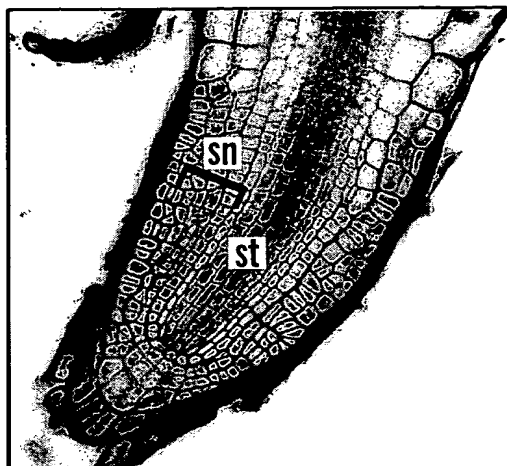
FIG. 8
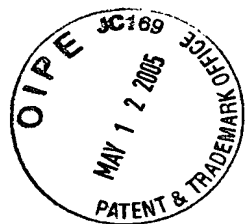
MDTLFRLVSL QQQQQSDSII TNQSSLSRTS TTTTGSPQTA YHYNFPQNDV VEECFNFFMD

EEDLSSSSSH HNHHNHNNPN TYYSPFTTPT QYHPATSSTP SSTAAAAALA SPYSSSGHHN

DPSAFSIPQT PPSFDFSANA KWADSVLLEA ARAFSDKDTA RAQQILWTLN ELSSPYGDTE

QKLASYFLQA LFNRMTGSGE RCYRTMVTAA ATEKTCSFES TRKTVLKFQE VSPWATFGHV

AANGAILEAV DGEAKIHIVD ISSTFCTQWP TLLEALATRS DDTPHLRLTT VVVANKFVND

QTASHRMMKE IGNRMEKFAR LMGVPFKFNI IHHVGDLSEF DLNELDVKPD EVLAINCVGA

MHGIASRGSP RDAVISSFRR LRPRIVTVVE EEADLVGEEE GGFDDEFLRG FGECLRWFRV

CFESWEESFP RTSNERLMLE RAAGRAIVDL VACEPSDSTE RRETARKWSR RMRNSGFGAV

GYSDEVADDV RALLRRYKEG VWSMVQCPDA AGIFLCWRDQ PVVWASAWRP T

FIG. 9
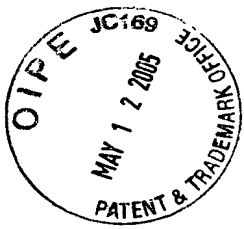
```
   1 aaaaaaaaaa aatggatact ctctttagac tagtcagtct ccaacaacaa caacaatccg
  61 atagtatcat tacaaatcaa tcttcgttaa gcagaacttc caccaccact actggctctc
 121 cacaaactgc ttatcactac aactttccac aaaacgacgt cgtcgaagaa tgcttcaact
 181 ttttcatgga tgaagaagac ctttcctctt cttcttctca ccacaaccat cacaaccaca
 241 acaatcctaa tacttactac tctcctttca ctactcccac ccaataccat cccgccacat
 301 catcaacccc ttcctccacc gccgcagccg cagctttagc ctcgccttac tcctcctccg
 361 gccaccataa tgacccttcc gcgttctcca tacctcaaac tcctccgtcc ttcgacttct
 421 cagccaatgc caagtgggca gactcggtcc ttcttgaagc ggcacgtgcc ttctccgaca
 481 aagacactgc acgtgcgcaa caaatcctat ggacgctcaa cgagctctct tctccgtaat
 541 gaaaaccgct tcattttcct tgtatttgtc tgaggttagg attagaccat tggttgttac
 601 tttcgaattc ttccaattta gttgttactt tcgaattctt ccatctctta gtttactaaa
 661 acaaacttat gtgccccata tttctccaac aatttgttga gtggtagctt acgttttact
 721 gtatacgctt ttgcaggtta tatcagcaca accattaatg atggcccggg atgtttgatg
 781 ctaagatgtc ctgacccatc ttgtcttgct gctgttggtc atgatatggt tgacaaatta
 841 gcgtctgaag acgaaaagga gaagtacaac agatattttc ttaggtctta tattgaagac
 901 aacagaaagg taagcagtct agaaaattta tatcacacag actggtatta atgtcgctgg
 961 tcttttattg agcaaaaact ggcttcttac ttcctccaag ctctcttcaa ccgcatgacc
1021 ggttcaggcg aacgatgcta ccgaaccatg gtaacagctg cagccacaga gaagacttgc
1081 tccttcgagt caacgcgaaa aactgtacta aagttccaag aagttagccc ctgggccacg
1141 tttggacacg tggcggcaaa cggagcaatc ttggaagcag tagacggaga ggcaaagatc
1201 cacatcgttg acataagctc cacgttttgc actcaatggc cgactcttct agaagcttta
1261 gccacaagat cagacgacac gcctcaccta aggctaacca cagttgtcgt ggccaacaag
1321 tttgtcaacg atcaaacggc gtcgcatcgg atgatgaaag agatcggaaa ccgaatggag
1381 aaattcgcta ggcttatggg agttcctttc aaatttaaca ttattcatca cgttggagat
1441 ttatctgagt ttgatctcaa cgaactcgac gttaaaccag acgaagtctt ggccattaac
1501 tgcgtaggcg cgatgcatgg gatcgcttca cgtggaagcc ctagagacgc tgtgatatcg
1561 agtttccgac ggttaagacc gaggattgtg acggtcgtag aagaagaagc tgatcttgtc
1621 ggagaagaag aaggtggctt tgatgatgag ttcttgagag ggtttggaga atgtttacga
1681 tggtttaggg tttgcttcga gtcatgggaa gagagttttc caaggacgag caacgagagg
1741 ttgatgctag agcgtgcagc gggacgtgcg atcgttgatc ttgtggcttg tgagccgtcg
1801 gattccacgg agaggcgaga gacagcgagg aagtggtcga ggaggatgag gaatagtggg
1861 tttggagcgg tggggtatag tgatgaggtg gcggatgatg tcagagcttt gttgaggaga
1921 tataaagaag gtgtttggtc gatggtacag tgtcctgatg ccgccggaat attcctttgt
1981 tggagagatc agccggtggt ttgggctagt gcgtggcggc caacgtaaag ggttgttttt
2041 attttttcat aaggaattc
```

FIG. 10
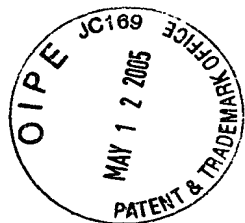
FIG. 11

2.5-kb SHORT-ROOT PROMOTER SEQUENCE

```
                 10          20          30          40          50
          1234567890  1234567890  1234567890  1234567890  1234567890
AGAAGCAGAG  CGTGGGGTTT  CTTCTAATAA  TTGTAGAAGA  AACTGATCAT      50
GAGAACATTT  GATCTACCAG  AGATGGTGAT  GACTCATAAG  ATGTAAATAT     100
CTACTGCATT  ATGTCTAGCC  TAGGCTATAA  TGTAGATTTG  ATCACTTTCT     150
TCATTAATTA  GTTTGGAATT  TTAGCATGAT  ATAGCATATA  TCTAAATATG     200
TCCGAAACTT  TCCTACATAC  TAGAAAATAT  GGAGAGTTAT  GTAATGTAGG     250
TTTGCTTGTT  AATATACAAA  ATAACATCAT  CATTTAGTTT  TTAGATTTTT     300
TATTTTATTT  TTTATAATGG  TGCTACGTAC  GTGGCGATCA  AATTATTCCA     350
ATTTTGAGAC  TTCGGGATTT  TAAACGAAAT  TAAACAATGG  GCATGAGCTC     400
GGGGGGATAG  ACAAGATTAA  TGCTTTGTAT  CGAGACAAAC  GAGAAAATCA     450
TGATGAGCCT  ATGCATTAAG  TGCCGTTGGT  TAATTAGAGG  TTCGCATATA     500
CATAAACCAG  TAGACATATG  GATAAATATG  AACACACACA  CCAAAAAAGT     550
GGGAAATCTA  AATAAGTGTA  GAGAATAATA  AGTCCTCAGG  TGGGAGATTC     600
AAAGAGAGGA  CAATGAAGGG  TATATAGACT  CTAAACAAAA  ATGGCATGAC     650
TTAGTGGAGA  GGGTTTTAAA  TTGAAACAAG  TAGGATTGAA  GAACAAGAAA     700
ACAAAGAAGC  ATGCCCTAGA  TTTCTGAGAT  AATAATTACA  CATTGCTGTT     750
TATATAAGGT  AAGAGAATAT  GACACATTGG  TTGGTTTCTT  ACGGGTAAAT     800
GTGAAGAAAA  AAAAATAGTA  ATATTTGAGA  AAATCTAAAA  TAGTAAAGAG     850
GTATATATGG  AGAAGAAGAG  AGAAAAGGGA  AAAATAGTGG  CAGAGAATGG     900
AGAGAGGTTA  GGAGGCAAAG  GCAAATGTGG  AGCTTTGATG  ATGTTGATGC     950
ACGCCGTCAG  CTTTTCTTCA  CGCCTGCTCC  CACTCACTCA  CACCTATGAA    1000
CATTCTCTCT  CTATTTTATA  ATTATATTCA  CATGTCTCTA  TGTTACTATG    1050
TAAATGGTGA  CCACTTAAGT  ATTTATATAT  CATGTATATA  TCTTATAGGT    1100
ATCATACAAA  ATGGTCATGA  AACTTTTGCA  ATTTCAATCT  ACTTGTTCAT    1150
TGTAGATGCT  AGCTTTTCAC  ATGTTTTGAA  AATTAGTCTG  GATCTGAAAT    1200
TCTTTAATTA  GCATTGTTTT  GTTGGTCAAC  GTTTAATTTC  TTGATTATTG    1250
ATGTCAAAAA  TTCAGAGCGT  TCAGAACTCT  TACACTAATT  TCTTAAAAAT    1300
AATCGATTAA  GAGAAAATAG  AGTTTTCATG  CACCAGTGTT  GATAGTAACG    1350
TAGTCGCGGA  ATGTCTAAAA  CGATTATGAG  TTTGGTGTTT  TGATTGGTTA    1400
GAATTGGTAT  TAGTAGGACA  TTCTAACTTT  TTTGTTAGTC  TGTTGATTTA    1450
GGATGCGTAA  AGAGTCTTTT  TATTTTACAC  CAGTTGAGAC  TTGGGATCGA    1500
TAGTACTTGA  AACACTTGGT  TGGTTTCATG  TATTTGGCCT  ATATATAAAC    1550
AAACATCGTA  ATTATATACG  GATTTTTTTC  GGAATTTTAC  GCCATATCTG    1600
TAAGTATATA  TAACATGCAT  GTCGTTTTCA  AATTCATATG  ATGAACGATC    1650
CACGTAAGTG  CTACTACTCC  TACAATATTG  CATGAGAGAG  ATATGTATTT    1700
ATAAATTTTA  TTTTGAAGAA  GAAATAAGAG  GGAAGGTTAC  TTGGGTGGAT    1750
CGATGTGAAA  ACAAAGAAG   AAAAAGCGAA  ACCCACTAAG  CCATTACATG    1800
ATATCGACCT  TCTTATCTTT  TTCCTCTTTA  TTTTATTTTT  CTCAGGACTT    1850
TTTTCTACTT  AATGAAACCT  CCAAACTATC  TAACTAATAC  ACTCCCATGT    1900
AGAATAAAGA  AAATTATATA  AGATATTGTT  GATATTTTGT  AACTAGAAAA    1950
TATATTTGCT  CTGTAATTTT  TCGTAAGTTA  AATCAACATT  TTTCAGTAGA    2000
AACAAATATT  ACTGCAAAAA  GTAGGATCAT  TATTTTTGTC  CAAAATCTCA    2050
GTTAGCTATA  GGGTTGTAGT  AAAAACAAAA  CACATTCTTG  ATTTGCCCCA    2100
AAAAATAAAG  AGAGAGAAGA  ATATTGTTCA  AAAGTGGTCT  CTTCTCTCTC    2150
TAATTATGTT  TTCACTAAAC  CCAATTAGAT  TCAAACAGTC  TACAAAGTCC    2200
AAAAGATAAA  CATGGGACAA  CAATTCGATG  CAAAAAATCC  TCTTTTCATG    2250
CTCTTTTTTT  ATTCTCTAGT  CTTTTAAATT  ACTAATAAAA  ACTCACAAAT    2300
CCACCAAACC  CATTCTCTAC  AACTCACCTT  CATCTAGATT  TACCCACTCC    2350
CACCGAGAAA  CACAAGAAAA  AAAATATACA  TATATAAATA  TACAAGACAA    2400
CACATGATGC  TGATGCAATA  TACACAACAA  AGTATTAAAT  CTTAGATATT    2450
GTGGGTCTCC  CTTTCTTCTA  TTCATTTTCT  TATTCATTAA  AAAAAAAAAA    2500
TG                                                           2502
```
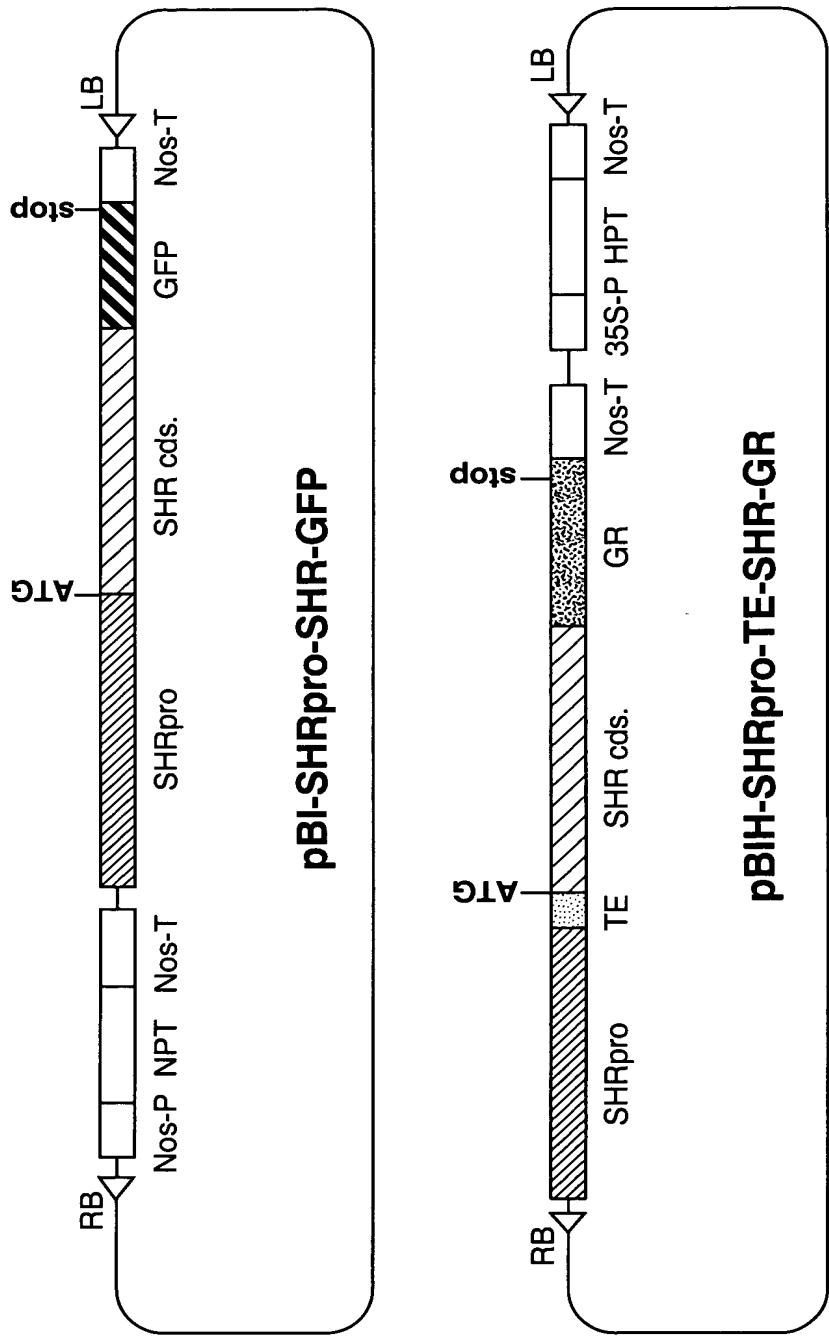
RB, right border sequence from *Agrobacterium* Ti plasmid
SHRpro, 2.5-Kb 5' upstream region of *SHORT-ROOT* gene
TE, translational enhancer element of tobacco etch virus
SHR cds., *SHORT-ROOT* protein coding region
GR, rat glucocorticoid receptor domain coding sequence
GFP, green fluorescent protein coding sequence
Nos-T, transcriprion terminator of nopaline synthetase gene
35S-P, cauliflower mosaic virus 35S promoter
HPT, hygromycin phosphotransferase coding sequence
NPT, neomycin phosphotransferase coding sequence
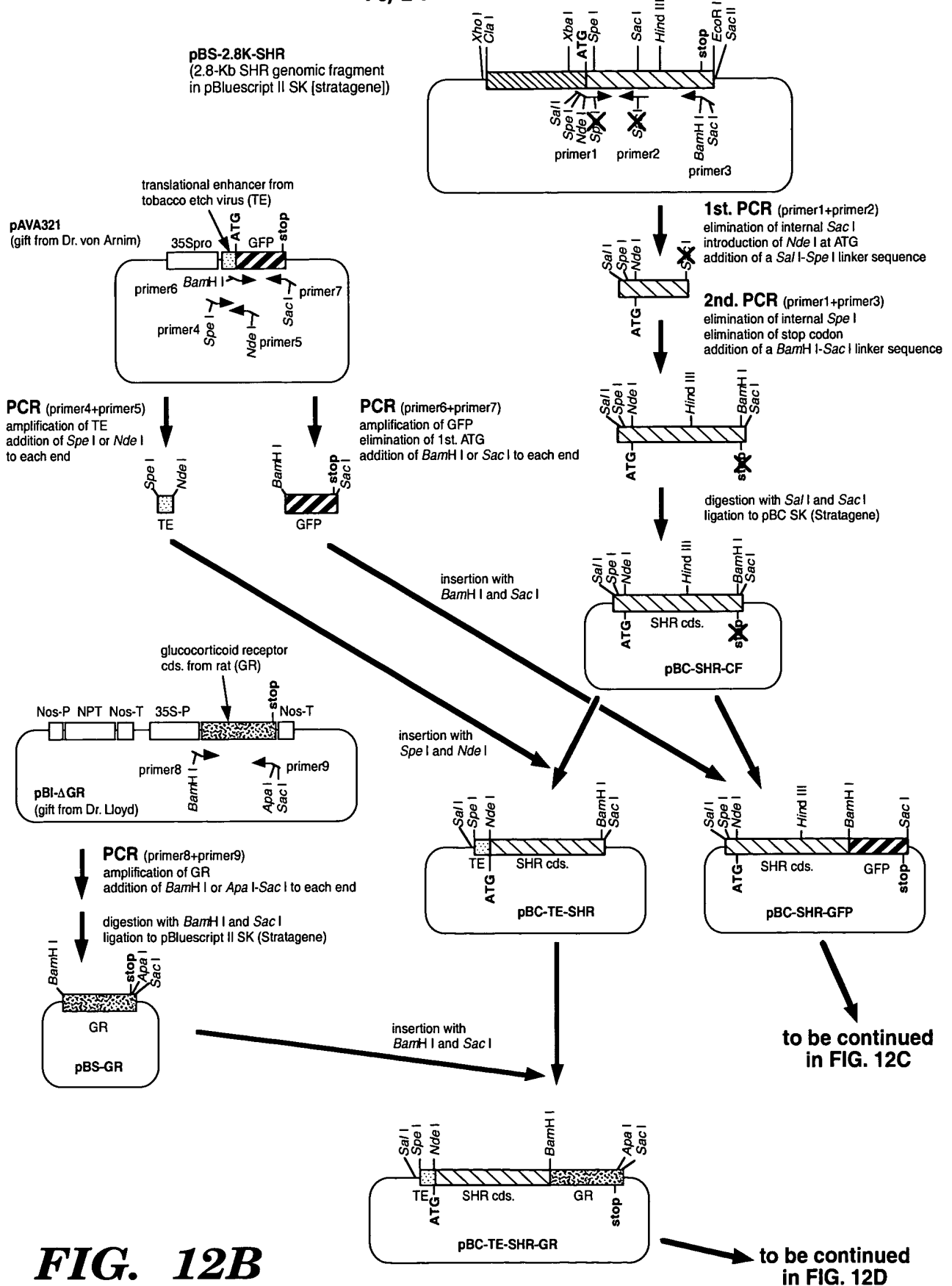**LB**, left border sequence from *Agrobacterium* Ti plasmid

pBI-SHRpro-SHR-GFP

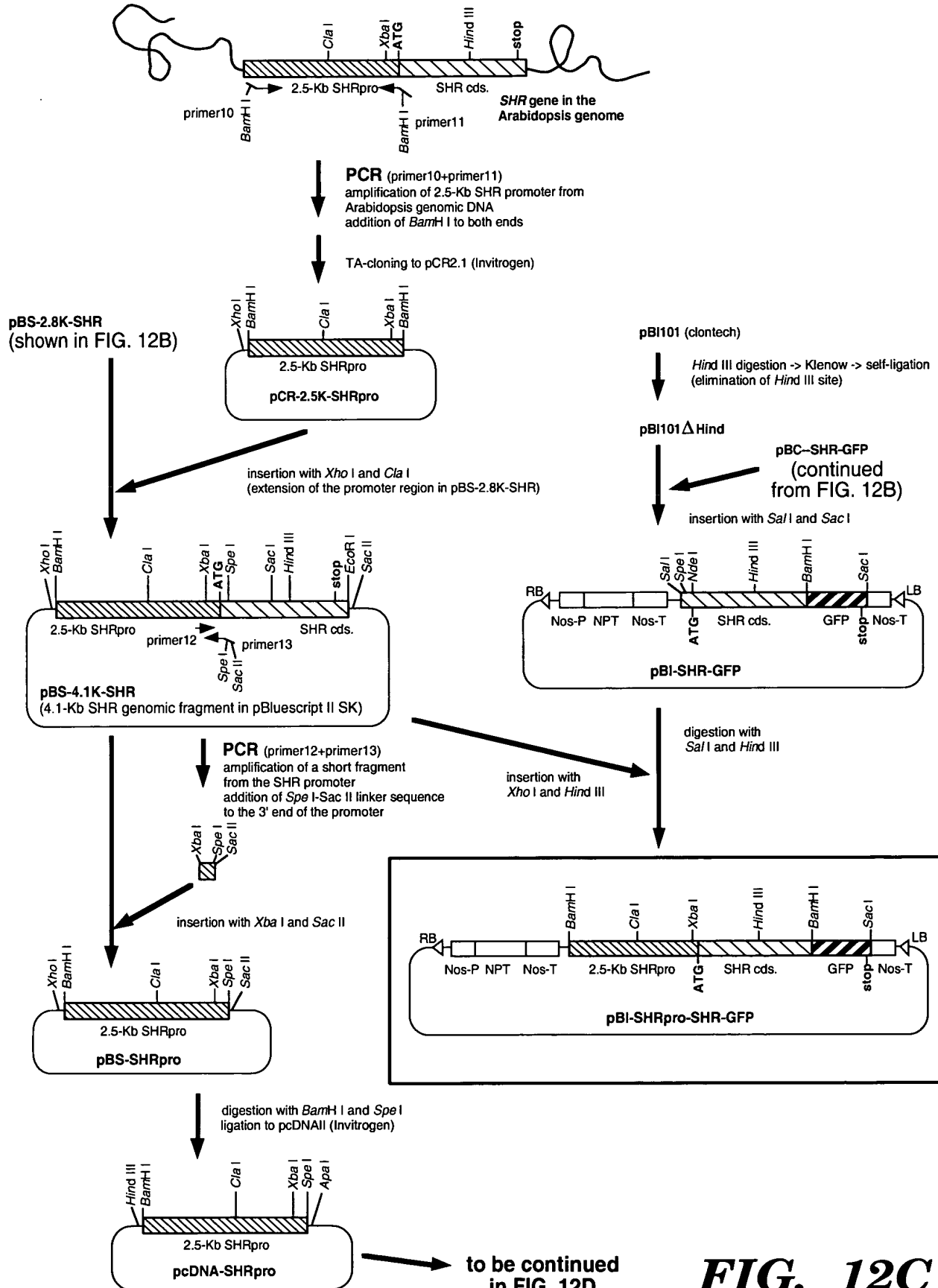pBIH-SHRpro-TE-SHR-GR

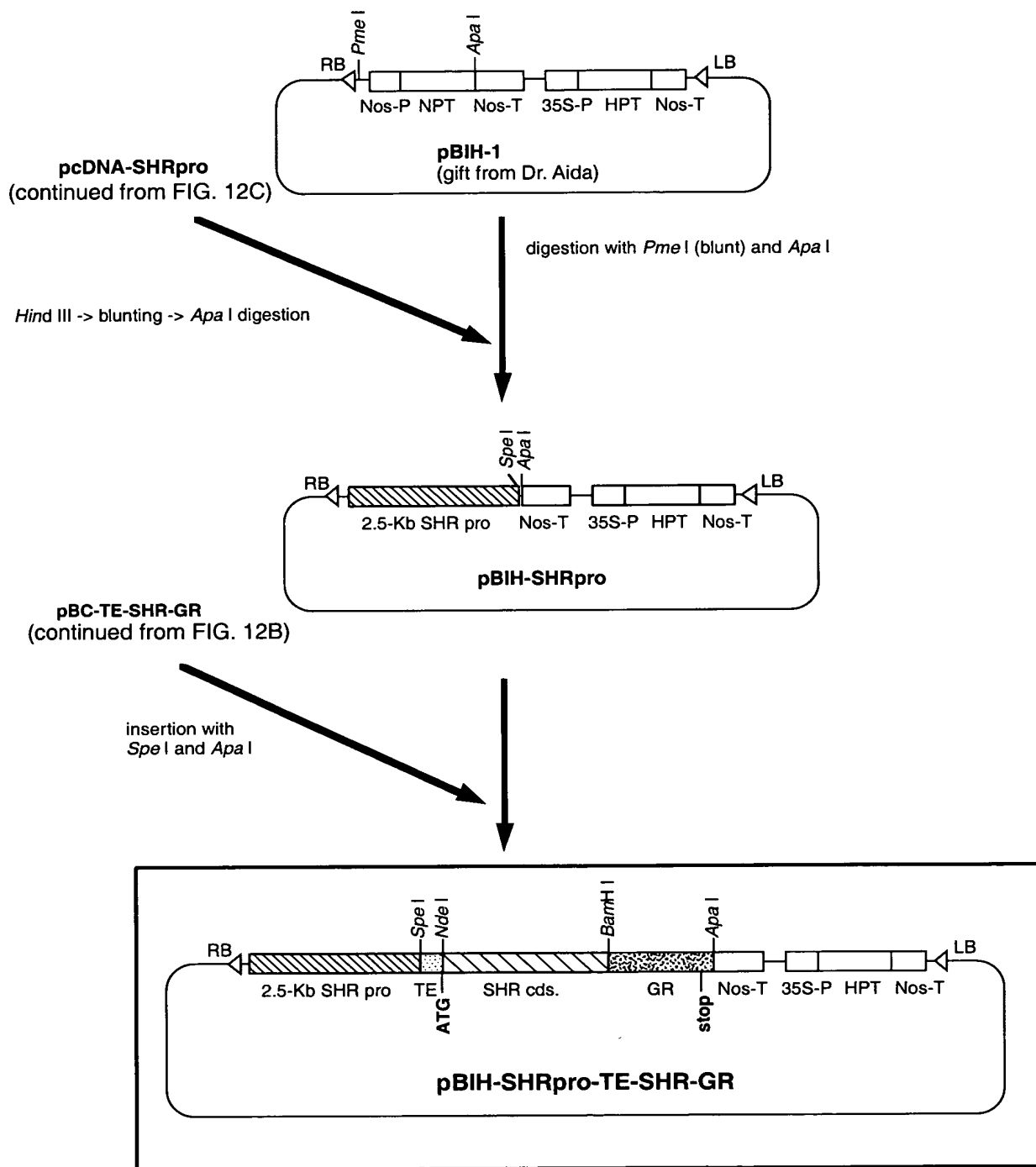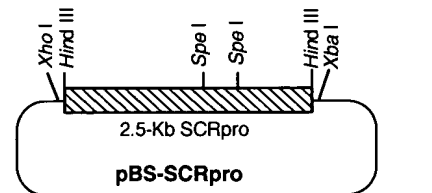*FIG. 12A*

18/24

pBS-2.8K-SHR
(2.8-Kb SHR genomic fragment
in pBluescript II SK [stratagene])
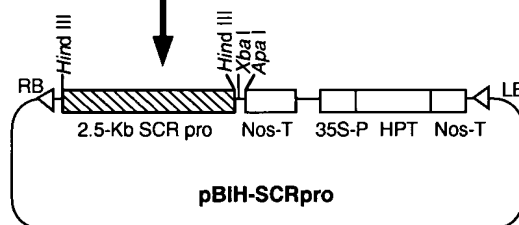
Xho I, *Cla* I, *Xba* I, *ATG*, *Spe* I, *Sac* I, *Hind* III, stop, *EcoR* I, *Sac*II

Sal I, *Spe* I, *Nde* I, *Sp* I — primer1   primer2   *BamH* I, *Sac* I — primer3
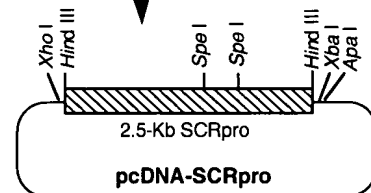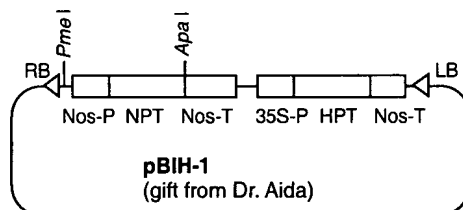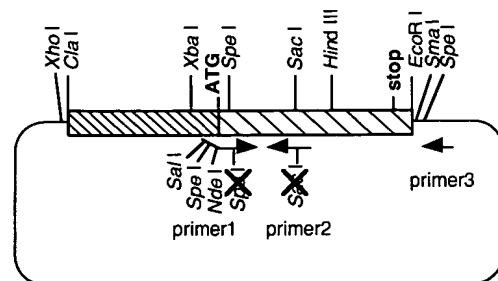
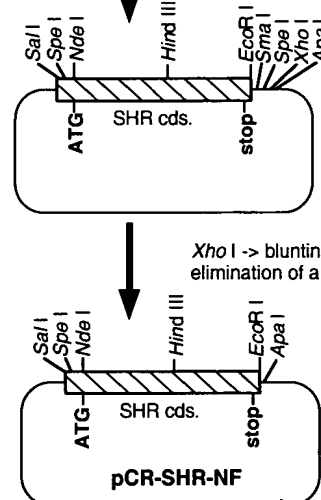**1st. PCR** (primer1+primer2)
elimination of internal *Sac* I
introduction of *Nde* I at ATG
addition of a *Sal* I-*Spe* I linker sequence

2nd. PCR (primer1+primer3)
elimination of internal *Spe* I
elimination of stop codon
addition of a *BamH* I-*Sac* I linker sequence

pAVA321
(gift from Dr. von Arnim)

35Spro   ATG   GFP   stop

primer6   *BamH* I   primer7
primer4   *Spe* I   *Nde* I   primer5
        Sac I

PCR (primer4+primer5)
amplification of TE
addition of *Spe* I or *Nde* I
to each end

translational enhancer from
tobacco etch virus (TE)

PCR (primer6+primer7)
amplification of GFP
elimination of 1st. ATG
addition of *BamH* I or *Sac* I to each end

Spe I   *Nde* I   *BamH* I   stop   *Sac* I
TE   GFP

Sal I, *Spe* I, *Nde* I, *Hind* III, *BamH* I, *Sac* I
ATG   stop

digestion with *Sal* I and *Sac* I
ligation to pBC SK (Stratagene)

insertion with
BamH I and *Sac* I

Sal I, *Spe* I, *Nde* I, *Hind* III, *BamH* I, *Sac* I
ATG   SHR cds.   stop
pBC-SHR-CF

glucocorticoid receptor
cds. from rat (GR)

Nos-P   NPT   Nos-T   35S-P   stop   Nos-T

primer8   *BamH* I   *Apa* I   primer9
                        Sac I

pBI-ΔGR
(gift from Dr. Lloyd)

insertion with
Spe I and *Nde* I

Sal I, *Spe* I, *Nde* I   *BamH* I, *Sac* I
TE   SHR cds.
ATG
pBC-TE-SHR

Sal I, *Spe* I, *Nde* I, *Hind* III, *BamH* I, *Sac* I
ATG   SHR cds.   GFP   stop
pBC-SHR-GFP

PCR (primer8+primer9)
amplification of GR
addition of *BamH* I or *Apa* I-*Sac* I to each end

digestion with *BamH* I and *Sac* I
ligation to pBluescript II SK (Stratagene)

BamH I   stop   *Apa* I   *Sac* I
GR
pBS-GR

insertion with
BamH I and *Sac* I

**to be continued
in FIG. 12C**

Sal I, *Spe* I, *Nde* I   *BamH* I   *Apa* I, *Sac* I
TE   SHR cds.   GR
ATG   stop
pBC-TE-SHR-GR

FIG. 12B

**to be continued
in FIG. 12D**

FIG. 12C

FIG. 12D

FIG. 13

SCRpro::SHR

WT

35Spro::SHR
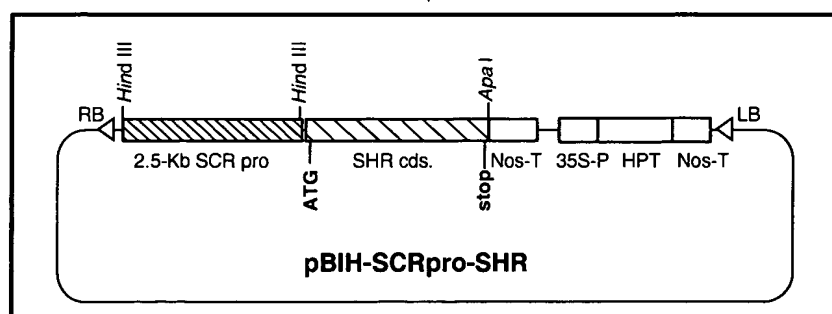
ECTOPIC *SHR* EXPRESSION CAUSED ABNORMAL ROOT CELL DIVISIONS

FIG. 14
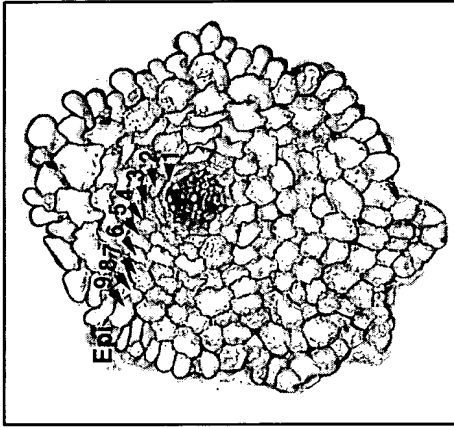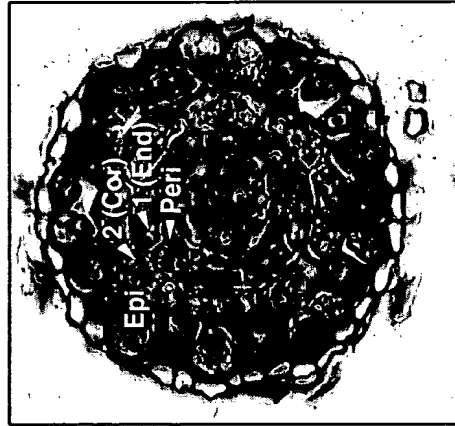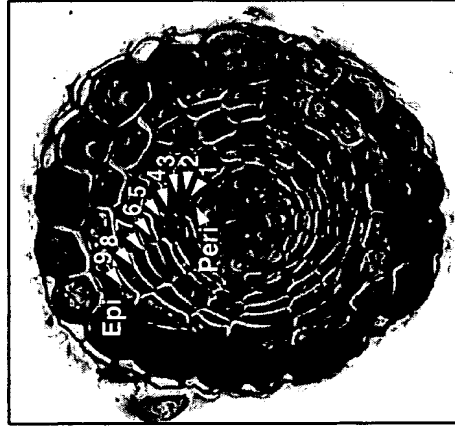
FIG. 15
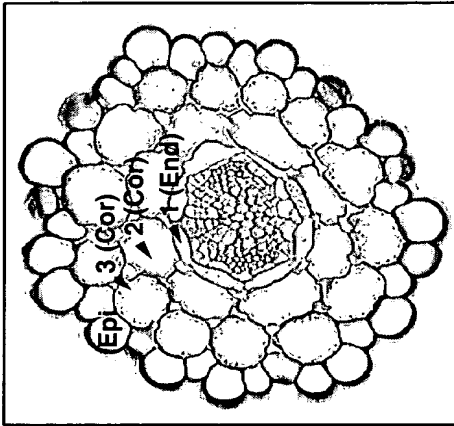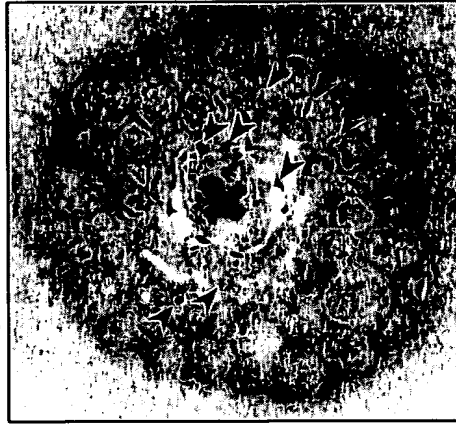
ECTOPIC *SHR* EXPRESSION UNDER THE *SCR* PROMOTER RESULTED IN THE INDETERMINATE CELL DIVISIONS IN GROUND TISSUE.
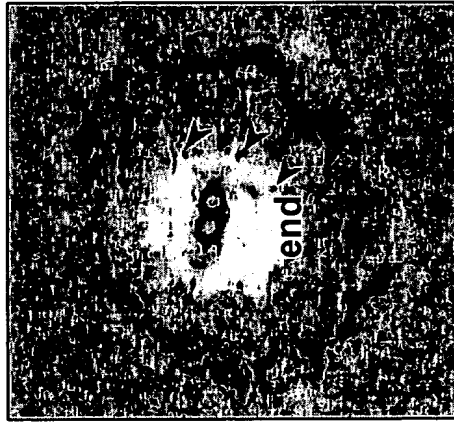
SCRpro::SHR transgenic

WT

Casparian strip occurs ectopically in the *SCRpro::SHR* transgenic root

FIG. 16